

NATIONAL TOXICOLOGY PROGRAM
Technical Report Series
No. 436



TOXICOLOGY AND CARCINOGENESIS

STUDIES OF *t*-BUTYL ALCOHOL

(CAS NO. 75-65-0)

IN F344/N RATS AND B6C3F₁ MICE

(DRINKING WATER STUDIES)

U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES
Public Health Service
National Institutes of Health

FOREWORD

The National Toxicology Program (NTP) is made up of four charter agencies of the U.S. Department of Health and Human Services (DHHS): the National Cancer Institute (NCI), National Institutes of Health; the National Institute of Environmental Health Sciences (NIEHS), National Institutes of Health; the National Center for Toxicological Research (NCTR), Food and Drug Administration; and the National Institute for Occupational Safety and Health (NIOSH), Centers for Disease Control. In July 1981, the Carcinogenesis Bioassay Testing Program, NCI, was transferred to the NIEHS. The NTP coordinates the relevant programs, staff, and resources from these Public Health Service agencies relating to basic and applied research and to biological assay development and validation.

The NTP develops, evaluates, and disseminates scientific information about potentially toxic and hazardous chemicals. This knowledge is used for protecting the health of the American people and for the primary prevention of disease.

The studies described in this Technical Report were performed under the direction of the NIEHS and were conducted in compliance with NTP laboratory health and safety requirements and must meet or exceed all applicable federal, state, and local health and safety regulations. Animal care and use were in accordance with the Public Health Service Policy on Humane Care and Use of Animals. The prechronic and chronic studies were conducted in compliance with Food and Drug Administration (FDA) Good Laboratory Practice Regulations, and all aspects of the chronic studies were subjected to retrospective quality assurance audits before being presented for public review.

These studies are designed and conducted to characterize and evaluate the toxicologic potential, including carcinogenic activity, of selected chemicals in laboratory animals (usually two species, rats and mice). Chemicals selected for NTP toxicology and carcinogenesis studies are chosen primarily on the bases of human exposure, level of production, and chemical structure. Selection *per se* is not an indicator of a chemical's carcinogenic potential.

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NTP TECHNICAL REPORT
ON THE
TOXICOLOGY AND CARCINOGENESIS
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CONTRIBUTORS

National Toxicology Program

Evaluated and interpreted results and reported findings

C.J. Alden, Ph.D.
G.A. Boorman, D.V.M., Ph.D.
D.A. Bridge, B.S.
J.R. Bucher, Ph.D.
J.D. Cirvello, B.S.
M.R. Elwell, D.V.M., Ph.D.
T.J. Goehl, Ph.D.
J.K. Haseman, Ph.D.
A.E. Radovsky, D.V.M., Ph.D.
G.N. Rao, D.V.M., Ph.D.
G.S. Travlos, D.V.M.
D.B. Walters, Ph.D.
K.L. Witt, M.S., Oak Ridge Associated Universities

Southern Research Institute

Conducted 13-week and 2-year drinking water studies, evaluated pathology findings

J.D. Prejean, Ph.D., Principal Investigator
D.R. Farnell, Ph.D., D.V.M.
J.E. Heath, D.V.M.
C. Lindamood, III, Ph.D.

Experimental Pathology Laboratories, Inc.

Provided pathology quality assurance

J.F. Hardisty, D.V.M., Principal Investigator
E.T. Gaillard, D.V.M., M.S.
B.F. Hamilton, D.V.M., Ph.D.

Dynamac Corporation

Prepared quality assurance audits

S. Brecher, Ph.D., Principal Investigator

Biotechnical Services, Inc.

Prepared Technical Report

D.D. Lambright, Ph.D., Principal Investigator
S.R. Gunnels M.A.
L.M. Harper, B.S.
M.J. Nicholls, B.S.
K.L. Shaw, B.A.

NTP Pathology Working Group

Evaluated slides, prepared pathology report on rats (30 December 1991)

J.R. Leininger, D.V.M., Ph.D., Chair
Pathology Associates, Inc.

W.W. Carlton, D.V.M., Ph.D.
Purdue University

E.T. Gaillard, D.V.M., M.S.
Experimental Pathology Laboratories, Inc.

R.A. Herbert, D.V.M., Ph.D.
National Toxicology Program

J.R. Hailey, D.V.M.
National Toxicology Program

M.P. Jokinen, D.V.M.
National Toxicology Program

R.R. Maronpot, D.V.M.
National Toxicology Program

C.C. Shackelford, D.V.M., M.S., Ph.D.
National Toxicology Program

Evaluated slides, prepared pathology report on mice (19 December 1991)

P.K. Hildebrandt, D.V.M., Chair
PATHCO, Inc.

J. Cullen, V.M.D., Ph.D.
North Carolina State University

J.R. Hailey, D.V.M.
National Toxicology Program

B.F. Hamilton, D.V.M., Ph.D.
Experimental Pathology Laboratories, Inc.

R.A. Herbert, D.V.M., Ph.D.
National Toxicology Program

M.P. Jokinen, D.V.M.
National Toxicology Program

C.C. Shackelford, D.V.M., M.S., Ph.D.
National Toxicology Program

G. Sykes, V.M.D.
Biosphere Pathology Services, Inc.

Evaluated slides, prepared pathology report on rat kidney step sections (extended evaluations) (3 September 1992)

J.R. Leininger, D.V.M., Ph.D., Chair
Pathology Associates, Inc.

S.L. Eustis, D.V.M., Ph.D.
National Toxicology Program

J.R. Hailey, D.V.M.
National Toxicology Program

R.A. Herbert, D.V.M., Ph.D.
National Toxicology Program

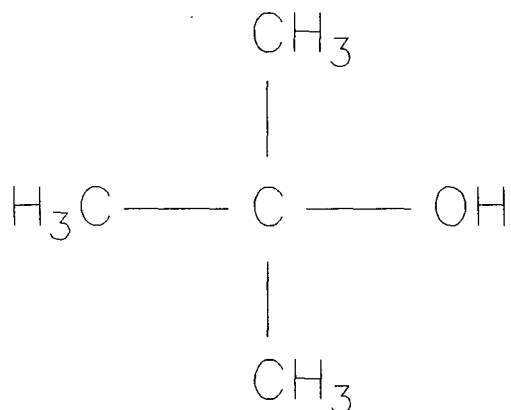
R.C. Sills, D.V.M., Ph.D.
National Toxicology Program

S. Stefanski, D.V.M., Ph.D.
North Carolina State University

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ABSTRACT



t-BUTYL ALCOHOL

CAS No. 75-65-0

Chemical Formula: $\text{C}_4\text{H}_{10}\text{O}$ Molecular Weight: 74.12

Synonyms: 2-Methyl-2-propanol, 2-methylpropan-2-ol, TBA, *t*-butanol, tertiary butyl alcohol, *t*-butyl hydroxide, trimethyl carbinol, trimethyl methanol

t-Butyl alcohol is widely used in the manufacture of perfumes and a variety of cosmetics. It is also used as a raw material in the production of isobutylene, which may be used to produce methyl tertiary butyl ether, a common gasoline additive, or to produce butyl elastomers used in the production of automobile tires. Male and female F344/N rats and B6C3F₁ mice were given *t*-butyl alcohol (greater than 99% pure) in drinking water for 13 weeks or 2 years. The genetic toxicity of *t*-butyl alcohol was assessed by testing the ability of the chemical to induce mutations in various strains of *Salmonella typhimurium* and in L5178Y mouse lymphoma cells, sister chromatid exchanges and chromosomal aberrations in cultured Chinese hamster ovary cells, and by measuring the frequency of micronucleated erythrocytes in mouse peripheral blood.

13-WEEK STUDY IN RATS

Groups of 10 male and 10 female F344/N rats were given 0, 2.5, 5, 10, 20, or 40 mg/mL *t*-butyl alcohol in drinking water for 13 weeks. All males and six

females given 40 mg/mL died during the study. Final mean body weights of 10 and 20 mg/mL males and of 40 mg/mL females were 12%, 17%, or 21% less than those of the corresponding controls, respectively. Serum sorbitol dehydrogenase activities in 10 and 20 mg/mL males were greater than that in the controls after 13 weeks. Serum alanine aminotransferase activity in 40 mg/mL females was greater than that in the controls after 2 weeks and greater in all exposed females after 13 weeks. Urine volumes of 10, 20, and 40 mg/mL males and females decreased, and urine specific gravity values increased. Transitional epithelial hyperplasia and inflammation of the urinary bladder were observed in 20 and 40 mg/mL males and 40 mg/mL females. Absolute and relative liver weights of all exposed groups of females and relative liver weights of 5, 10, and 20 mg/mL males were significantly greater than those of the controls. Absolute and relative kidney weights of all exposed groups of males and females were significantly greater than those of the controls. Incidences of mineralization of the kidney were significantly increased in 10, 20, and 40 mg/mL males. The severity of

nephropathy in 2.5, 5, 10, and 20 mg/mL males was significantly greater than that of the controls as was the accumulation of hyaline droplets in the kidney of 5, 10, and 20 mg/mL males. The incidences of nephropathy in 10, 20, and 40 mg/mL females were significantly greater than that of the controls.

13-WEEK STUDY IN MICE

Groups of 10 male and 10 female B6C3F₁ mice were given 0, 2.5, 5, 10, 20, or 40 mg/mL *t*-butyl alcohol in drinking water for 13 weeks. The deaths of two males and one female in the 40 mg/mL group were attributed to exposure to *t*-butyl alcohol. The final mean body weights of 20 and 40 mg/mL males and 40 mg/mL females were significantly lower than those of the controls. There were no biologically significant differences in hematology parameters of exposed and control groups of mice. Transitional epithelial hyperplasia and inflammation were observed in the urinary bladder of 20 and 40 mg/mL males and 40 mg/mL females.

2-YEAR STUDY IN RATS

Groups of 60 F344/N rats were given 0, 1.25, 2.5, or 5 mg/mL *t*-butyl alcohol (males) or 0, 2.5, 5, or 10 mg/mL *t*-butyl alcohol (females) in drinking water for 2 years. These correspond to average daily doses of approximately 90, 200, or 420 mg *t*-butyl alcohol/kg body weight for males and approximately 180, 330, or 650 mg *t*-butyl alcohol/kg body weight for females. Ten rats per group were evaluated after 15 months of chemical administration.

Survival, Body Weights, and Water Consumption

Survival rates of 5 mg/mL males and 10 mg/mL females were significantly lower than those of the controls. The final mean body weights of exposed groups of males were 15% to 24% lower than that of the controls, and the final mean body weight of 10 mg/mL females was 21% lower than that of the controls. Water consumption by males increased with dose; water consumption by females decreased with dose.

Hematology and Urinalysis

At the 15-month interim evaluation, there were no significant differences in hematology parameters in males and females, and there were no significant differences in urinalysis parameters in males. Females given 5 or 10 mg/mL had increased urine specific gravities and decreased urine volumes.

Pathology Findings

At the 15-month interim evaluation, relative kidney weights of 2.5 and 5 mg/mL males and absolute and relative kidney weights of 1.25, 2.5, and 5 mg/mL females were significantly greater than those of the controls. At 2 years, the incidence of mineralization in the kidney increased with dose and that of 5 mg/mL males was significantly greater than that of the controls. In the standard evaluation at the end of the study, the incidences of focal renal tubule hyperplasia and of adenoma were increased in exposed males and a carcinoma was observed in one 5 mg/mL male. Renal tubule hyperplasia occurred in one 10 mg/mL female. An extended evaluation of the kidney identified additional male rats with hyperplasia (control, 11/50; 1.25 mg/mL, 13/50; 2.5 mg/mL, 11/50; 5 mg/mL, 19/50) and renal tubule adenoma (7/50, 8/50, 15/50, 10/50); renal tubule carcinomas were identified in two 1.25 mg/mL males and in one 2.5 mg/mL male. Renal tubule adenoma was identified in one 5 mg/mL male from the 15-month extended evaluation. In the standard and extended evaluations combined, there were dose-related increased incidences of hyperplasia and adenoma. The severity of nephropathy and the incidence and severity of transitional cell hyperplasia of the kidney were increased in exposed male and female rats. Linear foci of mineralization were present in the renal papilla of exposed males.

2-YEAR STUDY IN MICE

Groups of 60 male and 60 female B6C3F₁ mice were given 0, 5, 10, or 20 mg/mL *t*-butyl alcohol in drinking water for 2 years. Exposure levels of 5, 10, or 20 mg/mL delivered average daily doses of approximately 540, 1,040, or 2,070 mg *t*-butyl alcohol/kg body weight to males and approximately 510, 1,020, or 2,110 mg/kg to females.

Survival, Body Weights, and Water Consumption

Survival of 20 mg/mL males was significantly lower than that of the controls. The final mean body weights of exposed groups of males were similar to those of the controls. The mean body weights of females given 20 mg/mL were 10% to 15% lower than those of the controls from week 13 to the end of the study. Water consumption by exposed groups of males and females was similar to that by the controls.

Pathology Findings

Incidences of thyroid gland follicular cell hyperplasia were significantly increased in all exposed groups of males and in 10 and 20 mg/mL females. The incidence of follicular cell adenoma or carcinoma (combined) was marginally increased in 10 mg/mL males (0 mg/mL, 1/60; 5 mg/mL, 0/59; 10 mg/mL, 4/59; 20 mg/mL, 2/57). The incidence of follicular cell adenoma was significantly increased in 20 mg/mL females (2/58, 3/60, 2/59, 9/59). The incidences of chronic inflammation and transitional epithelial hyperplasia of the urinary bladder were increased in 20 mg/mL males and to a lesser extent in 20 mg/mL females.

GENETIC TOXICOLOGY

t-Butyl alcohol was tested for induction of genetic damage *in vitro* and *in vivo*, and all results were negative. *In vitro*, t-butyl alcohol was negative in *Salmonella typhimurium* and mouse lymphoma cell mutation tests, and it did not induce sister chromatid

exchanges or chromosomal aberrations in cultured Chinese hamster ovary cells. These *in vitro* studies were conducted with and without metabolic activation (S9). *In vivo*, no increase in micronucleated erythrocytes was observed in peripheral blood samples from mice administered t-butyl alcohol in drinking water for 13 weeks.

CONCLUSIONS

Under the conditions of these 2-year drinking water studies, there was *some evidence of carcinogenic activity** of t-butyl alcohol in male F344/N rats based on increased incidences of renal tubule adenoma or carcinoma (combined). There was *no evidence of carcinogenic activity* in female F344/N rats receiving 2.5, 5, or 10 mg/mL t-butyl alcohol. There was *equivocal evidence of carcinogenic activity* of t-butyl alcohol in male B6C3F₁ mice based on the marginally increased incidences of follicular cell adenoma or carcinoma (combined) of the thyroid gland. There was *some evidence of carcinogenic activity* of t-butyl alcohol in female B6C3F₁ mice based on increased incidences of follicular cell adenoma of the thyroid gland.

Exposure to t-butyl alcohol was associated with mineralization and renal tubule hyperplasia in male rats, transitional epithelial hyperplasia and increased severity of nephropathy of the kidney in male and female rats, follicular cell hyperplasia of the thyroid gland in male and female mice, and chronic inflammation and hyperplasia of the urinary bladder in male mice and to a lesser extent in female mice.

* Explanation of Levels of Evidence of Carcinogenic Activity is on page 10. A summary of the Technical Reports Review Subcommittee comments and the public discussion on this Technical Report appears on page 12.

Summary of the 2-Year Carcinogenesis and Genetic Toxicology Studies of *t*-Butyl Alcohol

| | Male F344/N Rats | Female F344/N Rats | Male B6C3F ₁ Mice | Female B6C3F ₁ Mice |
|---|--|--|---|---|
| Doses | 0, 1.25, 2.5, or 5 mg/mL in drinking water [approximately 90, 200, or 420 mg/kg/day] | 0, 2.5, 5, or 10 mg/mL in drinking water [approximately 180, 330, or 650 mg/kg/day] | 0, 5, 10, or 20 mg/mL in drinking water [approximately 540, 1,040, or 2,070 mg/kg/day] | 0, 5, 10, or 20 mg/mL in drinking water [approximately 510, 1,020, or 2,110 mg/kg/day] |
| Body weights | Exposed groups lower than controls | 10 mg/mL group lower than controls | Exposed groups similar to controls | 20 mg/mL group lower than controls |
| 2-Year survival rates | 10/50, 6/50, 4/50, 1/50 | 28/50, 24/50, 22/50, 12/50 | 27/60, 36/60, 34/60, 17/60 | 36/60, 35/60, 41/60, 42/60 |
| Nonneoplastic effects | Kidney (standard evaluation): mineralization (26/50, 28/50, 35/50, 48/50); transitional epithelial hyperplasia (25/50, 32/50, 36/50, 40/50); severity of nephropathy (3.0, 3.1, 3.1, 3.3); (standard and extended evaluation): renal tubule hyperplasia (14/50, 20/50, 17/50, 25/50) | Kidney: transitional epithelial hyperplasia (0/50, 0/50, 3/50, 17/50); severity of nephropathy (1.6, 1.9, 2.3, 2.9) | Thyroid gland: follicular cell hyperplasia (5/60, 18/59, 15/59, 18/57) Urinary bladder: chronic inflammation (0/59, 3/59, 1/58, 37/59); transitional epithelial hyperplasia (1/59, 3/59, 1/58, 17/59) | Thyroid gland: follicular cell hyperplasia (19/58, 28/60, 33/59, 47/59) Urinary bladder: chronic inflammation (0/59, 0/60, 0/59, 4/57); transitional epithelial hyperplasia (0/59, 0/60, 0/59, 3/57) |
| Neoplastic effects | Kidney, renal tubule: (standard evaluation): adenoma (1/50, 3/50, 4/50, 2/50); adenoma or carcinoma (combined) (1/50, 3/50, 4/50, 3/50); (standard and extended evaluation): adenoma (8/50, 11/50, 19/50, 13/50); adenoma or carcinoma (combined) (8/50, 13/50, 19/50, 13/50) | None | None | Thyroid gland: follicular cell adenoma (2/58, 3/60, 2/59, 9/59) |
| Uncertain findings | None | None | Thyroid gland: follicular cell adenoma or carcinoma (combined) (1/60, 0/59, 4/59, 2/57) | None |
| Level of evidence of carcinogenic activity | Some evidence | No evidence | Equivocal evidence | Some evidence |

Summary of the 2-Year Carcinogenesis and Genetic Toxicology Studies of t-Butyl Alcohol (continued)

Genetic toxicology

| | |
|--|---|
| <i>Salmonella typhimurium</i> gene mutations: | Negative in strains TA98, TA100, TA1535, and TA1537 with and without S9 |
| Mouse lymphoma gene mutations: | Negative with and without S9 |
| Sister chromatid exchanges | |
| Cultured Chinese hamster ovary cells <i>in vitro</i> : | Negative with and without S9 |
| Chromosomal aberrations | |
| Cultured Chinese hamster ovary cells <i>in vitro</i> : | Negative with and without S9 |
| Micronucleated erythrocytes | |
| Mouse peripheral blood <i>in vivo</i> : | Negative |

EXPLANATION OF LEVELS OF EVIDENCE OF CARCINOGENIC ACTIVITY

The National Toxicology Program describes the results of individual experiments on a chemical agent and notes the strength of the evidence for conclusions regarding each study. Negative results, in which the study animals do not have a greater incidence of neoplasia than control animals, do not necessarily mean that a chemical is not a carcinogen, inasmuch as the experiments are conducted under a limited set of conditions. Positive results demonstrate that a chemical is carcinogenic for laboratory animals under the conditions of the study and indicate that exposure to the chemical has the potential for hazard to humans. Other organizations, such as the International Agency for Research on Cancer, assign a strength of evidence for conclusions based on an examination of all available evidence, including animal studies such as those conducted by the NTP, epidemiologic studies, and estimates of exposure. Thus, the actual determination of risk to humans from chemicals found to be carcinogenic in laboratory animals requires a wider analysis that extends beyond the purview of these studies.

Five categories of evidence of carcinogenic activity are used in the Technical Report series to summarize the strength of the evidence observed in each experiment: two categories for positive results (**clear evidence** and **some evidence**); one category for uncertain findings (**equivocal evidence**); one category for no observable effects (**no evidence**); and one category for experiments that cannot be evaluated because of major flaws (**inadequate study**). These categories of interpretative conclusions were first adopted in June 1983 and then revised in March 1986 for use in the Technical Report series to incorporate more specifically the concept of actual weight of evidence of carcinogenic activity. For each separate experiment (male rats, female rats, male mice, female mice), one of the following five categories is selected to describe the findings. These categories refer to the strength of the experimental evidence and not to potency or mechanism.

- **Clear evidence** of carcinogenic activity is demonstrated by studies that are interpreted as showing a dose-related (i) increase of malignant neoplasms, (ii) increase of a combination of malignant and benign neoplasms, or (iii) marked increase of benign neoplasms if there is an indication from this or other studies of the ability of such neoplasms to progress to malignancy.
- **Some evidence** of carcinogenic activity is demonstrated by studies that are interpreted as showing a chemical-related increased incidence of neoplasms (malignant, benign, or combined) in which the strength of the response is less than that required for clear evidence.
- **Equivocal evidence** of carcinogenic activity is demonstrated by studies that are interpreted as showing a marginal increase of neoplasms that may be chemical related.
- **No evidence** of carcinogenic activity is demonstrated by studies that are interpreted as showing no chemical-related increases in malignant or benign neoplasms.
- **Inadequate study** of carcinogenic activity is demonstrated by studies that, because of major qualitative or quantitative limitations, cannot be interpreted as valid for showing either the presence or absence of carcinogenic activity.

When a conclusion statement for a particular experiment is selected, consideration must be given to key factors that would extend the actual boundary of an individual category of evidence. Such consideration should allow for incorporation of scientific experience and current understanding of long-term carcinogenesis studies in laboratory animals, especially for those evaluations that may be on the borderline between two adjacent levels. These considerations should include:

- adequacy of the experimental design and conduct;
- occurrence of common versus uncommon neoplasia;
- progression (or lack thereof) from benign to malignant neoplasia as well as from preneoplastic to neoplastic lesions;
- some benign neoplasms have the capacity to regress but others (of the same morphologic type) progress. At present, it is impossible to identify the difference. Therefore, where progression is known to be a possibility, the most prudent course is to assume that benign neoplasms of those types have the potential to become malignant;
- combining benign and malignant neoplasm incidence known or thought to represent stages of progression in the same organ or tissue;
- latency in neoplasm induction;
- multiplicity in site-specific neoplasia;
- metastases;
- supporting information from proliferative lesions (hyperplasia) in the same site of neoplasia or in other experiments (same lesion in another sex or species);
- presence or absence of dose relationships;
- statistical significance of the observed neoplasm increase;
- concurrent control neoplasm incidence as well as the historical control rate and variability for a specific neoplasm;
- survival-adjusted analyses and false positive or false negative concerns;
- structure-activity correlations; and
- in some cases, genetic toxicology.

NATIONAL TOXICOLOGY PROGRAM BOARD OF SCIENTIFIC COUNSELORS TECHNICAL REPORTS REVIEW SUBCOMMITTEE

The members of the Technical Reports Review Subcommittee who evaluated the draft NTP Technical Report on *t*-butyl alcohol on June 21, 1994, are listed below. Subcommittee members serve as independent scientists, not as representatives of any institution, company, or governmental agency. In this capacity, subcommittee members have five major responsibilities in reviewing NTP studies:

- to ascertain that all relevant literature data have been adequately cited and interpreted,
- to determine if the design and conditions of the NTP studies were appropriate,
- to ensure that the Technical Report presents the experimental results and conclusions fully and clearly,
- to judge the significance of the experimental results by scientific criteria, and
- to assess the evaluation of the evidence of carcinogenic activity and other observed toxic responses.

Arnold L. Brown, M.D., Chair
University of Wisconsin Medical School
Madison, WI

Irma Russo, M.D., Principal Reviewer
Fox Chase Cancer Center
Philadelphia, PA

Paul T. Bailey, Ph.D.
Environmental and Health Sciences Laboratory
Mobil Oil Corporation
Princeton, NJ

Louise Ryan, Ph.D., Principal Reviewer
Division of Biostatistics
Harvard School of Public Health and
Dana-Farber Cancer Institute
Boston, MA

Meryl H. Karol, Ph.D.
Department of Environmental Occupational Health
University of Pittsburgh
Pittsburgh, PA

Robert E. Taylor, M.D., Ph.D.
Department of Pharmacology
Howard University College of Medicine
Washington, DC

Curtis D. Klaassen, Ph.D.
Department of Pharmacology and Toxicology
University of Kansas Medical Center
Kansas City, KS

Matthew J. van Zwieten, D.V.M., Ph.D.
Department of Safety Assessment
Merck Research Laboratories
West Point, PA

Claudia S. Miller, M.D.
University of Texas Health Sciences Center
San Antonio, TX

Mary Jo Vodicnik, Ph.D.
Lilly MSG Development Center
Belgium

Janardan K. Reddy, M.D.
Department of Pathology
Northwestern University Medical School
Chicago, IL

Jerrold Ward, D.V.M., Ph.D., Principal Reviewer
National Cancer Institute
Frederick, MD

SUMMARY OF TECHNICAL REPORTS REVIEW SUBCOMMITTEE COMMENTS

On June 21, 1994, the draft Technical Report on the toxicology and carcinogenesis studies of *t*-butyl alcohol received public review by the National Toxicology Program Board of Scientific Counselors Technical Reports Review Subcommittee. The review was held at the National Institute of Environmental Health Sciences, Research Triangle Park, NC.

Mr. J.D. Cirvello, NIEHS, introduced the toxicology and carcinogenesis studies of *t*-butyl alcohol by discussing the uses of the chemical and the routes of human exposure to the chemical, describing the experimental design, reporting on survival and body weight effects, and commenting on compound-related neoplasms and nonneoplastic lesions in rats and mice. Because of increased incidences of rare proliferative lesions of the renal tubule in exposed male rats, additional step sections from the kidneys of all control and exposed male rats were prepared and evaluated. The proposed conclusions were *some evidence of carcinogenic activity of t-butyl alcohol in male F344/N rats, no evidence of carcinogenic activity of t-butyl alcohol in female F344/N rats, equivocal evidence of carcinogenic activity of t-butyl alcohol in male B6C3F₁ mice, and some evidence of carcinogenic activity of t-butyl alcohol in female B6C3F₁ mice.*

Dr. Ward, a principal reviewer, disagreed with the proposed conclusion for male rats. He commented that with the standard pathology protocol, there were no significantly increased incidences of renal neoplasms or renal tubule hyperplasia, so no evidence of carcinogenic activity would be the appropriate conclusion. With the step-section technique, the incidence of neoplasms increased significantly in only the 2.5 mg/mL group. Thus, he considered the correct conclusion, based on the extended evaluation in male rats, to be equivocal evidence of carcinogenic activity. Mr. Cirvello responded that, based on the standard evaluation alone, equivocal evidence might be more appropriate. Only one adenoma occurred in controls and none occurred in controls from previous drinking water studies, while there were increased adenomas in all exposure groups in this study as well as a carcinoma in the 5 mg/mL group. Mr. Cirvello asked for discussion on the level of evidence based on the extended evaluation. Dr. J.K. Haseman, NIEHS, recommended that a formal statistical analysis of

neoplasm data from the extended evaluation be included in the report to clarify the chemical effect.

Dr. Ward said the pathology protocol should be more detailed and should include the number of step sections in the kidney, an explanation of the severity grading system for hyperplasias, and the number of lesions per section per rat. Dr. A. Radovsky, NIEHS, said the severity of hyperplasia is graded in terms of how closely the lesion resembles an adenoma (i.e., the size of the lesion and the extent of cellular atypia). Sixteen to 17 step sections per rat were taken instead of eight, which was standard in previous studies. She said this information would be added to the report.

Dr. Ryan, the second principal reviewer, questioned the proposed conclusions for male rats and male and female mice. With regard to male rats, she supported equivocal evidence of carcinogenic activity because none of the pairwise tests showed significant increases. Also, the life table trend test was significant but the logistic regression test was probably more relevant. For mice, she proposed no evidence for males and equivocal evidence for females. Although the incidence of thyroid adenomas in 20 mg/mL female mice was significantly greater than that of the controls, she felt this was not a clear exposure-related trend. Dr. Haseman stated that the incidence of combined renal neoplasms in the 2.5 mg/mL male rats was statistically significant by any test and the incidence in the 5 mg/mL group was significant with the addition of the one neoplasm from the interim sacrifice. Other factors supporting some evidence were neoplasm multiplicity at 2.5 mg/mL, increased hyperplasia at 5 mg/mL, and the occurrence of the uncommon carcinomas. With regard to the thyroid follicular cell adenomas in female mice, Dr. Haseman said this is a fairly uncommon neoplasm and the 15% incidence in the 20 mg/mL group was three times the highest incidence in drinking water controls and almost double the highest incidence in feed study controls. In addition, there were supporting increases in hyperplasia at 10 and 20 mg/mL. For male mice, he said that increased hyperplasias and similar but less impressive neoplasm findings than those in females suggested equivocal evidence.

Dr. Ryan asked why the results from 18-day and 13-week inhalation studies were not included in this Technical Report. Mr. Cirvello said that these studies will be reviewed and published separately in a toxicity study report. Dr. Ryan asked why renal tubule hyperplasia was considered rare when this lesion was observed in 12 male control rats in the extended evaluation. Dr. Radovsky responded that these lesions are uncommon in routine single sections of kidney from controls, but increasing the sample size to 16 sections per animal increased observations of this lesion.

Dr. Russo, the third principal reviewer, agreed with the proposed conclusions but had reservations about the proposed conclusion in male rats. She speculated that in this strain of rats, the chemical might be acting more as a promoter. Dr. J.R. Bucher, NIEHS, responded that the study was not designed as a promotion study, and further, the numbers of neoplasms observed in the extended evaluation would argue against a promotion effect. Dr. Russo said the report could benefit from some discussion of the lower susceptibility of female rats to the chemical and of the species-related organ specificity (i.e., kidney in rats versus thyroid in mice). Mr. Cirvello agreed to expand on this in the discussion.

There was more discussion on the merits and timing of extended pathologic evaluations. Dr. Haseman likened it to the difference between a partial evaluation and a definitive evaluation and said he would tend to give higher weight to the more definitive evaluation, which should be closer to the true neoplasms incidence. Dr. Russo urged more uniformity in the number of sections and the way they are taken in extended evaluations. Dr. Bucher commented that the NTP is careful to keep control data from standard histopathologic evaluations separate from data collected under step-section techniques. Dr. van Zwieten said step sections are most useful where an equivocal finding can be resolved through the extended evaluation. Dr. Haseman concurred.

Dr. Ward moved that the Technical Report on *t*-butyl alcohol be accepted with the revisions discussed and with the conclusions as written: *some evidence of carcinogenic activity of t-butyl alcohol in male F344/N rats, no evidence of carcinogenic activity of t-butyl alcohol in female F344/N rats, equivocal evidence of carcinogenic activity of t-butyl alcohol in male B6C3F₁ mice, and some evidence of carcinogenic activity of t-butyl alcohol in female B6C3F₁ mice.* Dr. van Zwieten seconded the motion, which was accepted unanimously with 11 votes.